A METHOD TO TEST THE HYPOTHESIS OF SELECTIVE FERTILIZATION.

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The hypothesis of selective fertilization has played an important rôle in several recent theories of sex determination and to some extent also in modern Mendelian speculations. It has been assumed for example that there are two classes of spermatozoa and two classes of eggs — male and female eggs and male and female sperm — and that fertilization is reciprocal in the sense that a male sperm can only fertilize a female egg and that a female sperm can only fertilize a male egg. On the other hand the more commonly accepted view is that any sperm can fertilize any egg.

Until this question is settled by direct observation or by experiment these two alternatives will continue to make uncertain our interpretations.

To put together one sperm and one egg would seem to be the simplest way to test the question. The operation may not in itself present insuperable difficulties but the chance of the spermatozoön reaching the surface of the egg is so small as to make the attempt rather hopeless; for there is no evidence to show that the spermatozoön is attracted towards the egg. The work of Buller, in particular, shows that accident alone determines the contact between the spermatozoa and the membranes, or the jelly, of the egg.

By means of the following simple method we have found it possible to study the problem of selective fertilization. We feel that while the number of cases here recorded is too small to settle so important a question, its application on a larger scale and on other animals should furnish conclusive evidence for or against selective fertilization.

We wish therefore at present to lay more emphasis on the possibilities of the method than on the certainty of demonstration from the number of recorded cases, and hope that others

may be led to study this important question by the same method.

We made use of the eggs and sperm of the mollusk *Cumingia*. The spermatozoa are quite large and can be readily seen with moderate magnification. The eggs are small so that the entire exposed surface can be watched closely. The eggs just laid were put into a drop of water on a slide, and a cover slip added. The cover slip was sufficiently supported so that the eggs were not too much compressed. It was advantageous to apply pressure because otherwise the spermatozoa may reach the egg above or below the horizon of observation; such sperm are lost to sight as a rule and their fate remains uncertain. Even with moderate pressure the spermatozoön sometimes slips in between the egg and the cover slip (above or below the horizon) and are lost to sight. In such cases further observation is worthless.

By means of a fine pointed pipette a small drop of water containing not too many spermatozoa was introduced at the edge of or under the cover slip. From the point of insertion the spermatozoa swim out in all directions and at some distance from the starting point the path of a single spermatozoön could be easily followed. The only way in which we were absolutely certain of seeing the first spermatozoön that reached the egg was to focus on an egg and wait until one came in contact with the egg.

It was seen that many spermatozoa swim past the egg and show no evidence of turning towards it, but those whose previous path was such that they ran into the jelly around the egg, bored into the jelly and often reached the surface of the egg. Whether after a spermatozoön has entered the jelly it ever turns towards the egg (if it did not have this orientation at the time of contact) is difficult to determine with certainty, but it is certain that spermatozoa may bore through without turning owards the egg. Some of the attached spermatozoa may show alternate periods of rest and activity, and in consequence change their position several times, and even end by entering the egg, but there is no evidence that one position is more directed than another.

The successful spermatozoa are those that strike the egg "head-on," and bore directly towards the surface. When the surface is reached the end of the spermatozoön appears to enter

the outermost layer of the egg. As a rule a pause follows, and it may take the spermatozoön from five to twenty minutes to disappear within the egg. The penetration is, as a rule, at first slow but later the spermatozoön may enter quite suddenly. Whether the sperm bores its way into the egg, or whether there is first a reaction between the surface of the egg and the spermatozoön so that the egg also takes a part in the process need not be discussed here, but it is important to note that no spermatozoa enter except those that stand with their long axis vertical to the surface and pierce the surface with the tip of the sperm head.

Our observations show that the first sperm that fulfills these conditions is received. In some of these cases a second sperm came in contact with the egg after the first had come in contact with it. In all such cases the first sperm only penetrated. There was no evidence in favor of selective fertilization, since in all forty cases the first sperm that approached in the normal position was the first to enter. It is highly improbable that forty times, this first sperm was the one suited to enter (assuming that two kinds exist) when there is no evidence that the eggs attract the sperm. Our general conclusion from the data here presented is that in this case selective fertilization does not occur, and since *Cumingia* is unisexual the temptation is to generalize this statement to include all such forms. Whether this extension is warrantable or not the fact remains that in this, the only case so far tested, the evidence is opposed to the hypothesis of selective fertilization.

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